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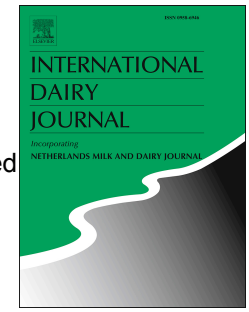

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Influence of protein standardisation media and heat treatment on viscosity and related physicochemical properties of skim milk concentrate

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ABSTRACT

The effects of heat treatment and protein standardisation on the physical properties of skim milk concentrates were determined. Protein standardisation was carried out by the addition of lactose or milk permeate to skim milk. Unstandardised and standardised skim milk was subjected to pasteurisation temperatures of 90 or 120 °C prior to evaporation whereafter the solids content was increased to 46% (w/w). Viscosity data showed non-standardised concentrates had the highest viscosity, followed by skim standardised with milk permeate and finally standardised with lactose. Thermal treatment at 120 °C also resulted in a higher viscosity than that at 90 °C for all concentrates. Particle size data of evaporated skim milk showed a bimodal size distribution for standardised skim milk with liquid milk permeate, compared with monomodal distribution profiles for unstandardised skim milk and lactose standardised skim milk. Overall, this study showed that protein standardisation and standardisation media significantly affected concentrate properties.

1. Introduction

Skim milk powder (SMP) is one of the most widely produced and commercially important commodity dairy powders and is used as an ingredient in a range of food applications. It is generally produced by evaporating and spray drying pasteurised skimmed milk. Heat treatment is carried out to decrease the microbial load and increase heat stability, which assists in achieving desired functional properties of the final ingredient, such as increasing acid gel strength in yoghurt manufacture (Damin, Alcantara, Nunes, & Oliviera, 2009; Lucey, Munro, & Singh, 2009).

The effects of thermal treatment on denaturation and aggregation of whey proteins in skim milk have previously been reported (Oldfield, Taylor, & Singh, 2005; Singh & Creamer, 1991; Singh & Newstead, 1992). During heating, β-lactoglobulin (β-lg) is irreversibly denatured, as its reactive thiol group is exposed, making it available to react with other free thiol groups (Jang & Swaisgood, 1990; Lowe et al., 2004; Vasbinder & de Kruif, 2003). Similarly, α-lactalbumin (α-la) is irreversibly denatured through disulphide bridging with β-lg (Mulvihill & Donovan, 1987). As caseins are non-globular proteins, limited heat-induced changes occur to their structure during thermal treatment, with the exception of κ-casein (Law, Horne, Banks, & Leaver, 1994). According to Jean, Renan, Famelart, and Guyomarc'h (2006), heat-induced aggregates in skim milk were essentially composed of disulphide-bonded κ-casein and denatured whey proteins. In fact, the denatured whey proteins can either interact with κ-casein in the serum phase or on the casein micelle surface depending on pH (Anema, 2007; Corredig & Dalgleish, 1996; Vasbinder & de Kruif, 2003). Therefore, following heat treatment, the protein profile of skim milk consists of native whey proteins, denatured and aggregated whey protein and casein-whey protein aggregates (Vasbinder, Alting, & de Kruif, 2003). Based on the level of heat-induced whey protein denaturation, SMP is typically classified into three categories: low, medium and high. The level of denatured whey protein is

generally used as an indicator of the heat classification for an SMP and is expressed as whey protein nitrogen index (WPNI) (Sikand, Tong, & Walker, 2008).

After heat treatment of skim milk, evaporation usually takes place to remove ~80% of the total water content prior to spray drying. During concentration as water is removed from the milk, viscosity increases due to higher solids content of the system; at the same time the distance between the protein molecules is reduced, therefore increasing the frequency of protein-protein interactions (Karlsson, Ipsen, Schrader, & Ardo, 2005). Structural changes in whey protein resulting from their denaturation also impact viscosity of the concentrate, where more extensive denaturation of whey protein results in greater protein-protein interactions (i.e., covalent and non-covalent bonds). Throughout lactation, the protein content of milk varies with time post-partum, therefore the protein content is often standardised to create a product with more consistent composition (and functionality) throughout the season. Protein standardisation may be carried out by the addition of lactose or milk permeate (in dry powder or reconstituted liquid format). Milk permeate is a co-product from the manufacture of milk protein concentrates and isolates from skim milk and contains all of the non-protein serum phase constituents of milk (i.e., lactose, minerals, vitamins and water). Their addition to skim milk allows for protein to be diluted to the desired level. While protein standardisation is regularly applied during the skim manufacturing process prior to heat treatment, the effect of protein standardisation on the viscosity of the skim milk when subsequently concentrated has not been fully addressed in detail to date.

The aim of this study was to determine the effects of heat treatment and protein standardisation (including standardisation media and mode of addition) on the rheological and physicochemical properties of skim milk concentrates following subsequent evaporation. A better understanding of multicomponent interactions taking place in the manufacturing process of SMP, and effects these have on the viscosity of SMP concentrate, can lead to optimisation, increased energy efficiency, less processing challenges (e.g., due to fouling) and overall

consistent quality of this valuable dairy ingredient.

2. Materials and methods

2.1. Materials

Skim milk was obtained from a local dairy processor and had a total solids level of 9.68% (w/w), with a protein content of 4.13% (w/w), equating to 42.7% (w/w) on a dry basis, and a casein:whey protein ratio ~77:22 using the Kjeldahl method (IDF, 2001). Lactose and milk permeate powders were also obtained from the same local dairy processor. The ash content of lactose and milk permeate powder were 0.3% (w/w) and 8.1% (w/w), respectively. The mineral composition of lactose and permeate powders were determined using inductively coupled plasma mass spectrometry (ICP-MS; Agilent Technologies, Santa Clara, CA, USA). Lactose and milk permeate powders contained 0.28 and 11.5% sodium, 0.94 and 21.5% potassium, 0.11 and 4.34% magnesium, 0.31 and 7.79% calcium, 0.06 and 2.94% chloride, 0.05 and 3.61% sulphate and 0.29 and 8.64% phosphate, respectively. All other chemicals and reagents used in the study were of analytical grade and sourced from Sigma-Aldrich (Arklow, Ireland).

2.2. Preparation of skim milk concentrates

Protein standardisation was carried out by addition of reconstituted lactose or milk permeate powders to the skim milk (protein content: 42.7%, w/w, dry basis) to give a final protein content of 34% (w/w; dry basis). Lactose and milk permeate powders were dissolved in warm water (60 °C) using a Silverson high shear mixer at a concentration of 9.9% (w/w) to maintain a constant solids level in the skim milk. The standardised skim milk (<8 °C) was

subsequently stirred gently for 10 min prior to thermal treatment. An additional trial was carried out in which the milk permeate powder was added directly to the skim milk via an in-line high shear mixer (YTRON-Z, 1.50FC, YTRON Process Technology GmbH, Bad Endorf, Germany) at <8 °C without any prior dilution in 60 °C water. All skim milk batches (50 kg) were then heat treated at 90 °C × 60 s (medium heat) or 120 °C × 270 s (high heat) using a MicroThermics Lab heat exchanger (MicroThermics, Raleigh, NC, USA) and evaporated to 46% total solids (TS) at 65 °C using a single-effect falling film evaporator (Anhydro F1 Lab; Copenhagen, Denmark) operating in recirculation mode. A summary of the trials carried out is shown in Table 1.

2.3. Whey protein nitrogen index

Whey protein nitrogen index (WPNI) was determined according to the GEA Niro method A 21a with results presented as mg native protein g^{-1} dry powder (mg g^{-1}). A WPNI (mg g^{-1}) value > 6 corresponds to low heat treatment, 1.5–6 corresponds to medium heat treatment and < 1.5 corresponds to high heat treatment (Sikand et al., 2008).

2.4. Gel electrophoresis of liquid skim milk

Protein profiles of skim milk were determined using pre-cast sodium dodecylsulphate polyacrylamide gel electrophoresis (SDS-PAGE) (Novex Technologies, ThermoFischer Scientific, Dublin, Ireland) under reducing and non-reducing conditions using the method described by Buggy, McManus, Brodkorb, McCarthy, and Fenelon (2017). After electrophoresis, the gels were stained overnight using 0.05% (w/v) Coomassie brilliant blue R-250 in 25% (v/v) isopropanol and 10% (v/v) acetic acid. After staining, the gels were de-stained using a 10% (v/v) isopropanol and 10% (v/v) acetic acid solution until a clear

background was achieved.

2.5. Titratable acidity and pH

Titratable acidity was measured for skim milk samples before and after heat treatment, and after evaporation using a method derived from that described by Dave and Shah (1997). Following evaporation, results were corrected for solids to be expressed at the same solids content as the untreated skim (9.68%). Skim milk (10 g) was titrated with sodium hydroxide (NaOH) using 1 mL of phenolphthalein as an indicator until a persistent faint pink colour appeared. The % of lactic acid was then calculated using the following equation:

$$\frac{\text{Titration result} \times (\text{mL of } 0.1 \text{ N NaOH}) \times 0.009 \times 100}{\text{Weight of sample (g)}} \quad (1)$$

The pH of skim milk samples was measured using a standard pH meter (Meterlab®, Radiometer Analytical, Villeurbanne, Lyon, France) at 20 °C.

2.6. Rheological measurements of skim milk concentrates

Apparent viscosity measurements of skim milk concentrates were carried out at 50 °C using a rotational rheometer (RST-CC TouchTM, Brookfield AMETEK, Middleboro, MA, USA), equipped with a coaxial cylinder and spindle, as described by O'Sullivan et al. (20 7). The shear rate was increased from 0 to 300 s^{-1} over 5 min, held at 300 s^{-1} for 2 min and decreased to 0 s^{-1} over 5 min. Measurements were immediately performed on the samples obtained after evaporation to avoid gelation and all analyses were performed in duplicate. The power law applied to the log-log plots of shear stress (τ) versus shear rate ($\dot{\gamma}$) was used to obtain the flow behaviour parameters, consistency coefficient (K) and flow behaviour index (n) as detailed by Anema, Lowe, Lee, and Klostermeyer (2014). The flow behaviour index values

were used to describe the flow behaviour of liquid samples, where $n < 1$, $n > 1$ and $n = 1$ indicate shear-thinning, shear-thickening and Newtonian flow behaviour, respectively.

2.7. Particle size measurements of liquid skim milk

The particle size distribution of skim milk before and after evaporation were measured by static light scattering (SLS) using a laser-light diffraction unit (Hydro MV, Mastersizer 3000, Malvern Instruments Ltd, Malvern, UK) equipped with a 300 RF lens. Particle and dispersant (i.e., water) refractive indices were set at 1.38 and 1.33, respectively. Size measurements were recorded as the median (D_{50}), volume mean ($D_{4,3}$) and cumulative diameters (D_{90} and D_{10}), while size distributions were obtained using polydisperse analysis. Measurements were recorded when the laser obscuration reached ~5%. All measurements were carried out in triplicate.

3. Results

3.1. WPNI and SDS-PAGE protein profiles of skim milk

WPNI values for unstandardised and standardised skim milk prior to heat treatment were in the range of 7.47 ± 0.00 to 7.68 ± 0.01 mg g^{-1}, and were typical of low heat skim milk (Sikand et al., 2008). WPNI values for skim milk after medium (90 °C × 60 s) and high (120 °C × 270 s) heat treatment were in the range of 3.35 ± 0.01 to 4.42 ± 0.04 mg g^{-1} and 0.75 ± 0.01 to 1.36 ± 0.02 mg g^{-1}, respectively, which correlate well with the standard heat classification system as described in section 2.4. After medium heat treatment, the lowest 174 WPNI value was obtained for unstandardised skim milk (WPNI of 3.35 ± 0.01 mg g^{-1}) compared with standardised skim milk with lactose (WPNI 4.00 ± 0.01 mg g^{-1}), liquid milk permeate (WPNI

4.42 mg g^{-1}) or milk permeate powder (WPNI 4.09 ± 0.00 mg g^{-1}). After high heat treatment conditions, unstandardised skim milk had a lower WPNI value (0.75 ± 0.01 178 mg g^{-1}) compared with skim milk standardised with lactose (1.29 mg g^{-1}) or added milk permeate powder (1.36 ± 0.02 mg g^{-1}). However, standardising with liquid milk permeate prior to heat treatment resulted in the lowest WPNI value of 0.42 ± 0.02 mg g^{-1}.

SDS-PAGE analysis carried out under reducing conditions showed all samples had similar protein profiles regardless of heat treatment (Fig. 1A). However, non-reducing SDS-PAGE (Fig. 1B) showed the effect of heat treatment on protein aggregation. Fig. 1B (lanes 9–12) showed milk samples which were not heat treated following pasteurisation, with β-lg, α-la and the larger molecular weight minor whey proteins clearly present. Skim milk heat treated at 90 °C × 60 s (Fig. 1 B; lanes 1–3) showed more faint bands for both β-lg and α-la, while many of the minor whey proteins were absent. Skim milk heat treated at 120 °C × 270 s (Fig. 1B; lanes 4–6) showed β-lg to be almost completely absent while there was a reduction in α-la band intensity. Increasing the severity of heat treatment from 90 °C × 60 s to 120 °C × 270 s resulted in a greater reduction in band intensity for each of the whey proteins and an increase in band intensity for aggregated material positioned in the stacking gel (Fig. 1B; lanes 4, 5 and 6).

3.2. Titratable acidity and pH

There was no significant ($P > 0.05$) difference in titratable acidity results for all samples prior to evaporation (Table 2). All treatments had an acid value in the range of 0.10–0.13% before evaporation. However, after evaporation unstandardised skim milk had higher acidity values, compared with protein standardised skim milks which had 0.16 and 0.15% acidity values for the medium and high heat treatments, respectively. Standardising the protein content of skim milk with either lactose or milk permeate resulted in lower values for titratable acidity. The acidity results are similar to those obtained by Dave and Shah (1997), who reported acidity

values of 0.14% for whole milk following heat treatment of 85 °C for 1800 s.

pH values of skim milk before and after heat treatment and after evaporation are shown in Table 2. Prior to heat treatment, skim milk standardised with milk permeate had higher pH compared with skim milk standardised with lactose. There was a significant ($P < 0.05$) decrease in pH for all skim milk samples after heat treatment with high heat samples having lower pH values compared with medium heat (Table 2). After evaporation the pH of all samples decreased to the range of 6.08 to 6.22.

3.3. Rheological properties of skim milk concentrates

Viscosity increased significantly ($P < 0.05$) with increasing severity of heat treatment (from 90 °C × 60 s to 120 °C × 270 s) for both unstandardised and standardised skim milk (Fig. 2A). Viscosity of high heat unstandardised skim (i.e., 42.7%, w/w, protein, dry basis) was higher than the viscosity of all other samples at 103 mPa s (Fig. 2A), followed by unstandardised skim milk after medium heat treatment (68.8 mPa s). Protein standardisation (i.e., 34%, w/w, protein, dry basis) resulted in a lower viscosity across all skim milk concentrates. The use of lactose for protein standardisation gave the lowest viscosity for medium and high heat skim milk (Fig. 2A; 24.9 and 28.0 mPa s, respectively). Protein standardisation with liquid milk permeate resulted in concentrates with the highest viscosity across all standardised milks (i.e., viscosities for medium and high heat samples of 38.4 and 43.6 mPa s, respectively). Power law analysis of shear stress versus shear rate is shown in Table 3 and Fig. 2B. In general, the shear stress increased with increasing shear rate, indicating the concentrates were non-Newtonian and shear-thinning (Fig. 2B), as also described by Anema et al. (2014) for skim milk concentrates at 45% TS. The flow behaviour index (n) values ranged from 0.56 to 0.81, with the higher level of protein in unstandardised skim milks resulting in lower n values and corresponding higher consistency coefficient values (Table 3).

Low flow behaviour index values (n = 0.56; Table 3) correlated well with high viscosity measurements for the high heat unstandardised concentrate (i.e., 103 mPa s; Fig. 2A).

3.4. Particle size distribution measurements

Particle size distribution (PSD) data of unheated skim milk is shown in Table 4 and Fig. 3A. Prior to heat treatment PSD profiles of unstandardised skim milk were monomodal ($D_{4,3}$ = 0.21 µm) and did not change with the addition of lactose (Table 4; Fig. 3A). Skim milk standardised by direct addition of milk permeate powder had a monomodal PSD profile (Fig. 3A) with a $D_{4,3}$ value of 0.58, which was larger than for unstandardised and standardised skim milk with lactose and significantly lower (P < 0.05) than milk standardised by liquid permeate addition ($D_{4,3}$ = 6.16 µm) (Table 4).

PSD profiles of heat treated skim milk measured after evaporation for unstandardised skim milk and skim milk standardised with lactose were similar for medium (Fig. 3B) and high heat treatment (Fig. 3C) conditions, with $D_{4,3}$ values ~0.2 µm (Table 4). Skim milk concentrates standardised with milk permeate had larger $D_{4,3}$ values with liquid milk permeate having the largest $D_{4,3}$ value. High heat treatment also resulted in higher $D_{4,3}$ values compared with medium heat for skim milk concentrates standardised with either liquid or powder milk permeate (Table 4). PSD profiles also showed a bimodal size distribution for skim standardised using liquid milk permeate (Fig. 3B and C; 5–30 µm).

4. Discussion

The viscosity of skim milk concentrate limits the solids content that can be achieved during evaporation prior to spray drying, due to reasons of evaporator fouling and reduced atomisation efficiency. This study showed the impact high protein concentrations can have on

viscosity, especially in unstandardised skim milks. It is important to note that the TS content achieved after evaporation (i.e., 46% TS) in this study was chosen so as to avoid skim milk gelation which occurs rapidly above 50% TS (Olivares, Achkar, & Zorrilla, 2016). The viscosity and WPNI of skim milk was most significantly affected by protein concentration followed by heat treatment, and finally by the type and mode of addition of the standardisation media. Overall, the viscosity of skim milk concentrate reduced with standardisation using either lactose or milk permeate. The higher viscosity values shown for unstandardised skim milk compared with standardised milks, even at medium heat treatment temperatures (Fig. 2A) can be attributed to the higher protein content (42.7%, w/w, dry basis) in the unstandardised skim milk. The higher level of protein in unstandardised milks resulted in a higher level of denatured and aggregated protein material, as shown by the lower WPNI values following medium and high heat treatments.

The degree to which viscosity was reduced was dependent on the standardisation material used and its mode of addition. For standardised skim milk samples, liquid milk permeate had the greatest effect on the viscosity, WPNI and PSD, followed by direct addition of milk permeate powder and finally liquid lactose having the least effect on these properties of the concentrates. The use of lactose for protein standardisation showed clear benefits for skim milk samples treated at both medium and high heat treatments. The difference observed in viscosity between skim milk with added lactose or milk permeate is likely due to the higher ash content in the milk permeate (8.1%, w/w), compared with lactose (0.3%, w/w).

The higher level of mineral addition during standardisation with milk permeate may lead to a higher ionic strength, reduction in electro-negative charges and increase in divalent bridging caused during heat treatment, all of which lead to more denatured/aggregated whey protein as shown by the low WPNI values in Section 3.1. A decrease in pH upon concentration was seen for all concentrates (Table 2) and may be attributed to the release of hydrogen ions and a concomitant solubilisation of calcium phosphate. Sutariya, Huppertz, and Patel (2017)

showed a pH drop of ~0.4 pH units after evaporation (50–55% TS) of skim milk, and is similar to data shown in Table 2 in this study. Bienvenue, Jimenez-Florez, and Singh (2003) stated that during evaporation/concentration, the increase in ionic strength reduces the electrostatic repulsion between casein micelles causing a collapse in the κ-casein layer leading to weakly flocculated casein micelles in skim milk concentrates. This corresponds to the rheological properties shown in Fig. 2B, where the skim milk concentrates demonstrated shear-thinning behaviour, indicating that high hydrodynamic forces disrupted weaker protein-protein interactions, such as ionic and hydrogen bonds at high shear rates.

The occurrence of weak protein-protein bonds may be supported by the presence of a bimodal size distribution in unheated skim milk with added liquid milk permeate (Fig. 3A). This may be due to the fact that when standardising using liquid milk permeate, there was a longer period of exposure of the protein to the minerals present in the permeate, compared with when adding permeate powder. Future work may entail measuring the particle size of unheated skim milk and their corresponding concentrates after exposure to high shear to determine if the aggregates present in skim milk standardised with liquid milk permeate are reversible. Therefore, maintaining a high rotational shear rate during concentrate storage prior to drying may prevent significant increases in viscosity.

5. Conclusion

The physicochemical properties of protein standardised skim milk concentrates were affected predominantly by protein content, heat treatment temperature and by the mode of addition and composition of the standardisation media (i.e., lactose or milk permeate). To obtain the lowest viscosity for skim milk at a given heat treatment temperature, reducing the protein content by standardisation is particularly effective; where standardisation using lactose resulted in a lower viscosity compared with milk permeate. However, where milk permeate

was used, its addition in powder form as opposed to liquid provides some benefits, particularly, a lower concentrate viscosity. Particle size distribution and whey protein denaturation of skim milk concentrates varied with protein content, type and mode of addition of standardisation media and heat treatment temperature. Dairy processing plants should acknowledge that these parameters will affect evaporation run times and achievable solids contents. The findings of this study provide useful information to dairy processors on optimisation of viscosity for increased efficiency during processing.

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Figure legends

Fig. 1. SDS-PAGE patterns of skim milk carried out under (A) reducing and (B) non-reducing conditions of: lane 1, medium heat unstandardised; lane 2, medium heat standardised with lactose; lane 3, medium heat standardised with liquid milk permeate; lane 4, high heat unstandardised; lane 5, high heat standardised with lactose; lane 6, high heat standardised with liquid milk permeate; lane 7, medium heat standardised with milk permeate powder; lane 8, high heat standardised with milk permeate powder; lane 9, non-heat treated unstandardised; lane 10, non-heat treated standardised with lactose; lane 11, non-heat treated standardised with liquid milk permeate; lane 12, non-heat treated standardised with milk permeate powder.

Fig. 2. Apparent viscosity (shear rate 300 s^{-1}; 50 °C) (A) and flow curves of log-log dependency of shear stress versus shear rate at 50 °C (B) of concentrated (46%, w/w, solids content) skim milk as a function of protein content, heat treatment (open symbols, medium heat treatment; closed symbols, high heat treatment) and standardisation media and mode: ○ and ●, unstandardised; ◇ and ◆, standardised with lactose; □ and ■, standardised with liquid permeate; △ and ▲, standardised with milk permeate powder.

Fig. 3. Particle size distribution profiles of unstandardised skim milk (**—**), skim milk standardised with lactose (**—**), skim milk standardised with liquid milk permeate (**•••**) and skim milk standardised with milk permeate powder (**•••**) prior to heat treatment (A), following medium heat treatment, 90 °C × 60 s and evaporation (B) and following high heat treatment, 120 °C × 270 s and evaporation (C).

Table 1

Processing conditions and sample codes for trials conducted.

Sample code	Heat treatment	Standardisation medium	Mode of addition
USM Med	90 °C × 60 s	n/a	n/a
USM High	120 °C × 270 s	n/a	n/a
Lact. Med	90 °C × 60 s	Lactose	Liquid
Lact. High	120 °C × 270 s	Lactose	Liquid
Liquid Perm. Med	90 °C × 60 s	Permeate	Liquid
Liquid Perm. High	120 °C × 270 s	Permeate	Liquid
Perm. Powder Med	90 °C × 60 s	Permeate	Powder
Perm. Powder High	120 °C × 270 s	Permeate	Powder

Table 2

Titratable acidity (% lactic acid) and pH of skim milk samples prior to heat treatment, post heat treatment and post evaporation. [a]

Treatment	Pre heat		Post heat		Post Evaporation	
	Acidity (%)	pH	Acidity (%)	pH	Acidity (%)	pH
USM Med	0.13	6.63	0.12	6.59	0.16	6.16
USM High	-	-	0.12	6.53	0.15	6.08
Lactose Med	0.10	6.72	0.10	6.61	0.12	6.18
Lactose High	-	-	0.10	6.57	0.11	6.16
Liquid Perm. Med	0.12	6.70	0.12	6.64	0.13	6.26
Liquid Perm. High	-	-	0.12	6.59	0.13	6.16
Perm. Powder Med	-	-	0.11	6.63	0.12	6.22
Perm. Powder High	-	-	0.11	6.53	0.13	6.20

[a] For treatment codes, refer to Table 1.

Table 3

Rheological properties of unstandardised and standardised skim milk (46%, w/w) concentrate. [a]

Treatment	Consistency coefficient, K (Pa s^n $\times 10^{-2}$)	Flow behaviour index, n
USM Med	45.5 ± 14.8^{b}	0.68 ± 0.06^{ab}
USM High	134 ± 22.8^{a}	0.56 ± 0.03^{a}
Lactose Med	9.30 ± 0.50^{b}	0.81 ± 0.01^{b}
Lactose High	10.3 ± 1.01^{b}	0.77 ± 0.02^{b}
Liquid Perm. Med	22.7 ± 10.1^{b}	0.70 ± 0.08^{ab}
Liquid Perm. High	19.4 ± 4.65^{b}	0.74 ± 0.05^{b}
Perm. Powder Med	12.7 ± 1.06^{b}	0.76 ± 0.04^{b}
Perm. Powder High	16.9 ± 1.90^{b}	0.74 ± 0.04^{b}

[a] For treatment codes, refer to Table 1; values within a column not sharing a common superscript differed significantly ($P < 0.05$).

Table 4

Particle size distribution and mean diameter ($D_{4,3}$) values (in μm) of unstandardised and standardised skim milk measured prior to heat treatment (9.68%, w/w) and after evaporation (46.0%, w/w). [a]

Treatment	D_{10}	D_{50}	D_{90}	$D_{4,3}$
Unheated				
USM	0.06 ± 0.00 [b]	0.17 ± 0.00 [a]	0.41 ± 0.00 [a]	0.21 ± 0.00 [a]
Lactose	0.06 ± 0.00 [b]	0.18 ± 0.01 [a]	0.41 ± 0.00 [a]	0.21 ± 0.00 [a]
Liquid Perm.	0.05 ± 0.00 [b]	0.21 ± 0.00 [a]	28.0 ± 1.97 [d]	6.16 ± 0.57 [e]
Perm. Powder	0.03 ± 0.00 [a]	0.14 ± 0.00 [a]	0.60 ± 0.00 [a]	0.58 ± 0.01 [a]
Medium Heat				
USM	0.07 ± 0.00 [b]	0.19 ± 0.00 [a]	0.42 ± 0.00 [a]	0.22 ± 0.00 [a]
Lactose	0.07 ± 0.00 [b]	0.19 ± 0.00 [a]	0.41 ± 0.00 [a]	0.22 ± 0.00 [a]
Liquid Perm.	0.03 ± 0.00 [a]	0.16 ± 0.00 [a]	8.16 ± 0.06 [b]	1.84 ± 0.01 [c]
Perm. Powder	0.03 ± 0.00 [a]	0.14 ± 0.00 [a]	0.70 ± 0.01 [a]	0.70 ± 0.01 [a]
High Heat				
USM	0.08 ± 0.00 [b]	0.21 ± 0.00 [a]	0.41 ± 0.00 [a]	0.23 ± 0.00 [a]
Lactose	0.08 ± 0.00 [b]	0.20 ± 0.00 [a]	0.41 ± 0.00 [a]	0.22 ± 0.00 [a]
Liquid Perm.	0.04 ± 0.00 [a]	0.18 ± 0.00 [a]	12.5 ± 0.06 [c]	2.67 ± 0.01 [d]
Perm. Powder	0.03 ± 0.00 [a]	0.16 ± 0.00 [a]	0.88 ± 0.01 [a]	1.09 ± 0.03 [b]

[a] Values within a column not sharing a common superscript differed significantly ($P < 0.05$).

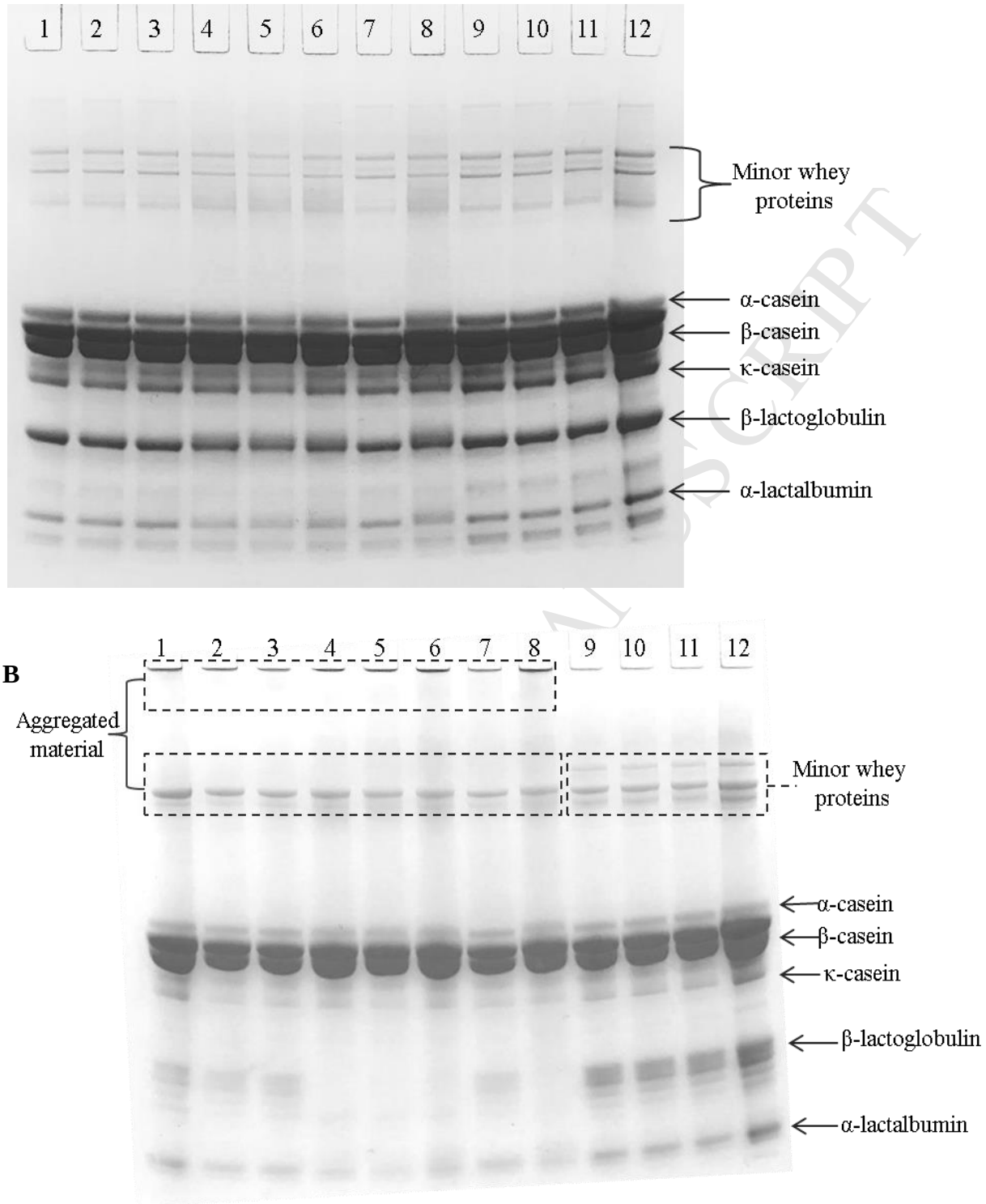

A
1 2 3 4 5 6 7 8 9 10 11 12
Minor whey proteins
α-casein
β-casein
κ-casein
β-lactoglobulin
α-lactalbumin
B
1 2 3 4 5 6 7 8 9 10 11 12
Aggregated material
Minor whey proteins
α-casein
β-casein
κ-casein
β-lactoglobulin
α-lactalbumin

Fig. 1.

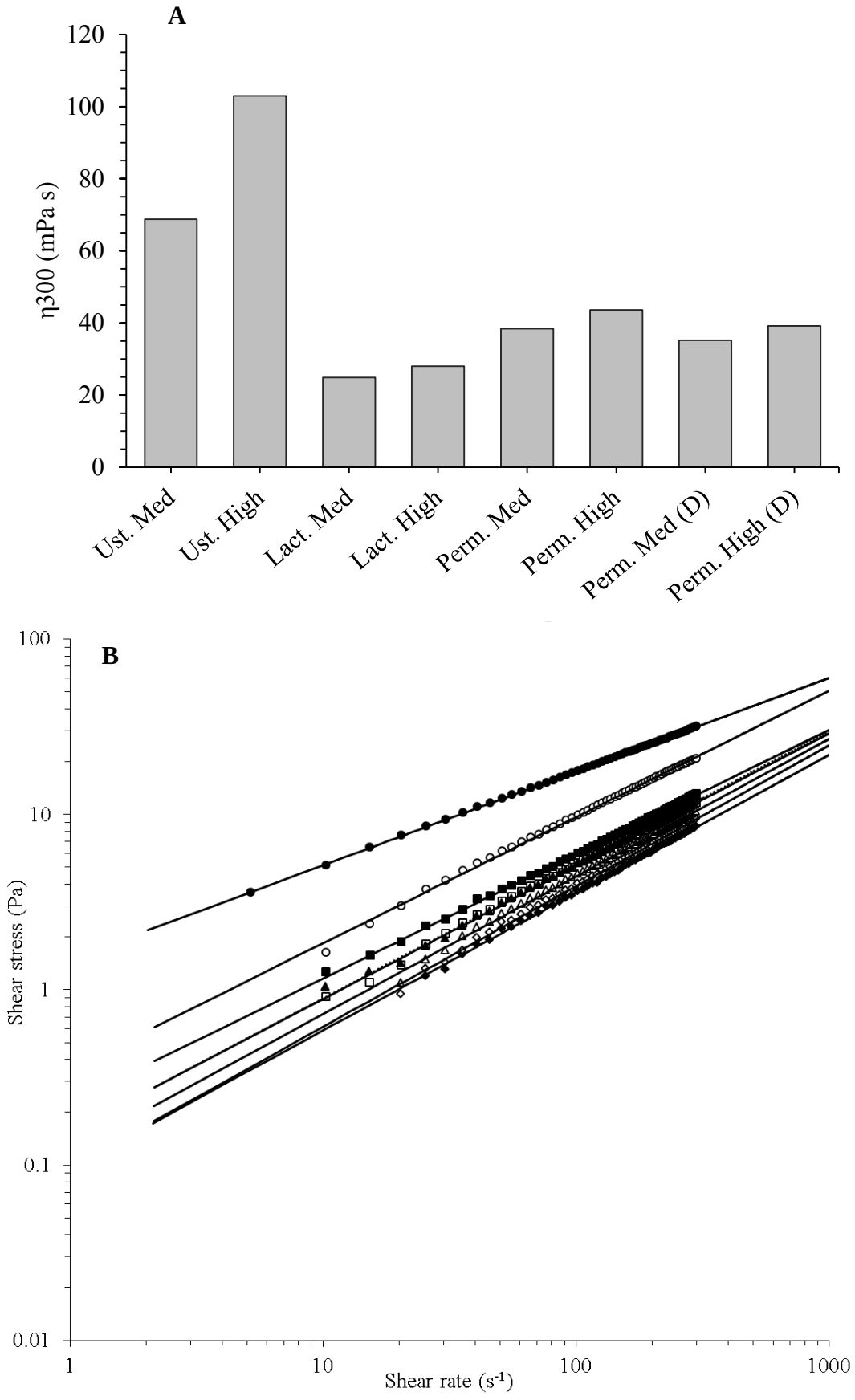
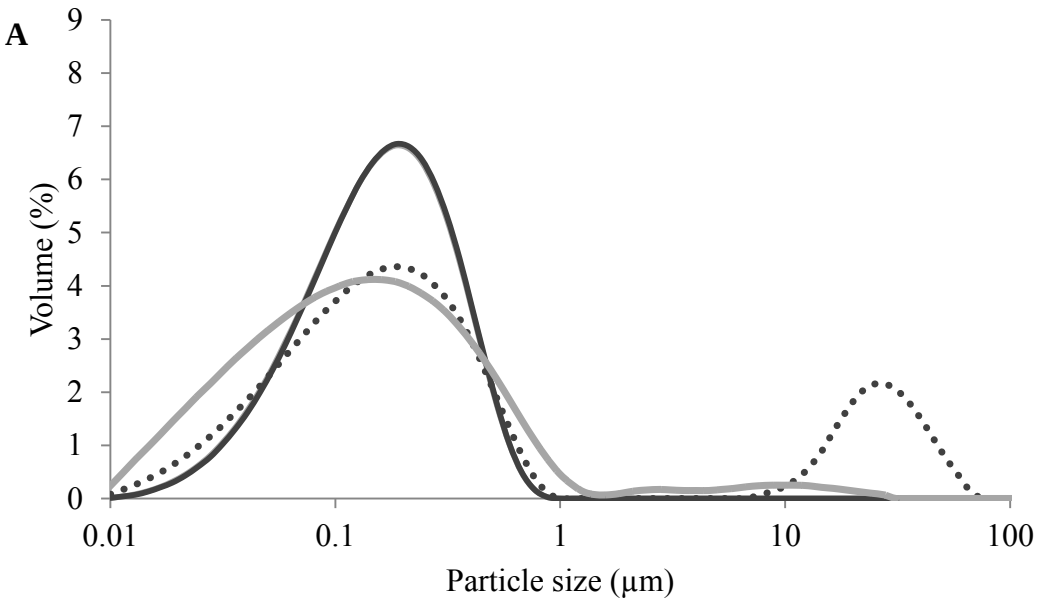
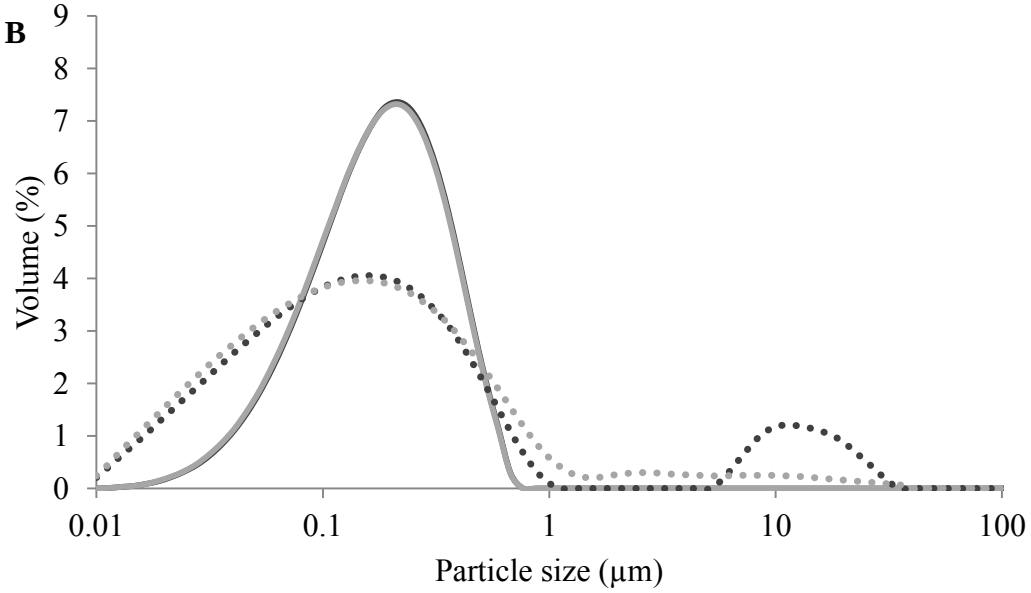

A
120
100
80
60
40
20
0
η300 (mPa s)
Ust. Med
Ust. High
Lact. Med
Lact. High
Perm. Med
Perm. High
Perm. Med (D)
Perm. High (D)
B
100
10
1
0.1
0.01
Shear stress (Pa)
1
10
100
1000
Shear rate (s-1)

Fig. 2.

16


A
Volume (%)
0
1
2
3
4
5
6
7
8
9
0.01
0.1
1
10
100
Particle size (μm)

17
18


B
Volume (%)
0
1
2
3
4
5
6
7
8
9
0.01
0.1
1
10
100
Particle size (μm)

19
20

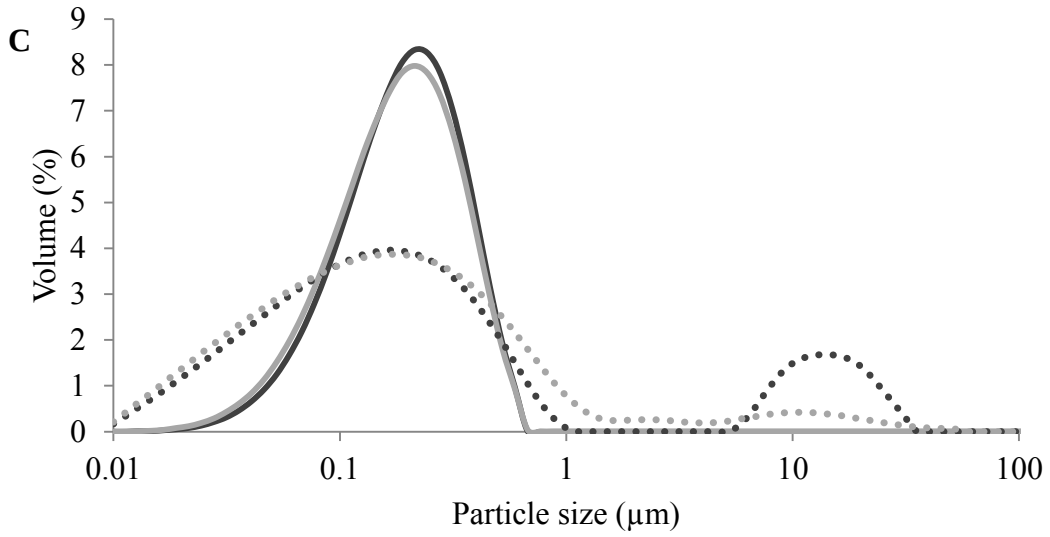


Fig. 3.